

What is claimed is:

1. A method for purifying one or more selected cells, comprising:

(a) contacting a plurality of cells immobilized in proximity to a capture matrix, the capture matrix capable of localizing a product secreted by one or more of the cells, with an agent that selectively binds to the product, the agent capable of generating a signal detectable as a property of light;

(b) illuminating a population of the cells, the population contained in a frame;

(c) detecting two or more properties of light directed from the frame, wherein a first property of light identifies substantially all cells of the population, and the second property of light identifies product localized to the capture matrix;

(d) locating (i) substantially all cells of the population with reference to the detected first property of light, and (ii) one or more selected cells with reference to the detected second property of light, and

(e) irradiating the non-selected cells, wherein each non-selected cell receives a substantially lethal dose of radiation,

whereby one or more selected cells having a desired product secretion profile are purified.

2. The method of claim 1, wherein the product is selected from a polypeptide, an antibody, an antibody fragment, a cytokine, a growth factor, an enzyme, a hormone, a neurotransmitter, a signaling molecule, and a therapeutic protein.

3. The method of claim 1, wherein the capture matrix comprises a substance selected from Protein G, Protein A, an antibody, an antibody fragment, an aptamer, and a ligand for the product.

4. The method of claim 1, wherein the capture matrix comprises a gel.

5. The method of claim 1, further comprising the step of (f) illuminating a further population of the cells, the further population contained in a further frame, and repeating steps (c) through (e).

6. The method of claim 1 or 5, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the population.

7. The method of claim 1 or 5, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the population.

8. The method of claim 1 or 5, wherein the one or more selected cells are cells that produce an undetectable level of product.

9. The method of claim 5, further comprising repeating step (f) followed by steps (c) through (e) until substantially all of the non-selected cells in the plurality of cells receive a substantially lethal dose of radiation.

10. The method of claim 9, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the plurality.

11. The method of claim 9, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the plurality.

12. The method of claim 9, wherein the one or more selected cells are cells that produce an undetectable level of product.

13. The method of claim 1, further comprising allowing the purified one or more selected cells to proliferate.

14. The method of claim 1, wherein steps (b) through (e) are automated.

15. The method of any of claims 1, 5 or 9, wherein the frame is a portion of a field-of-view, the field-of-view having an area selected from the group of greater than 5 mm<sup>2</sup>, greater than 10 mm<sup>2</sup>, greater than 20 mm<sup>2</sup>, greater than 40 mm<sup>2</sup>, greater than 80 mm<sup>2</sup>, and greater than 160 mm<sup>2</sup>.

16. The method of claim 1, wherein the agent that selectively binds to the product is selected from an antibody, an antibody fragment, an aptamer, a substrate, and a ligand.

17. The method of claim 1, wherein the one or more selected cells are identified with reference to a signal value corresponding to an amount of product localized to the capture matrix in the vicinity of the one or more selected cells.

18. The method of claim 1, wherein the substantially lethal dose of radiation is delivered to each non-selected cell in one or more radiation pulses.

19. The method of claim 1, wherein the substantially lethal dose of radiation has an energy density selected from the group of greater than  $0.1 \text{ J/cm}^2$ , greater than  $0.3 \text{ J/cm}^2$ , greater than  $1 \text{ J/cm}^2$ , greater than  $3 \text{ J/cm}^2$ , greater than  $10 \text{ J/cm}^2$ , greater than  $30 \text{ J/cm}^2$ , and greater than  $100 \text{ J/cm}^2$ .

20. The method of claim 1, wherein the substantially lethal dose of radiation has an irradiance selected from the group of greater than  $10^7 \text{ W/cm}^2$ , greater than  $10^8 \text{ W/cm}^2$ , greater than  $10^9 \text{ W/cm}^2$ , greater than  $10^{10} \text{ W/cm}^2$ , and greater than  $10^{11} \text{ W/cm}^2$ .

21. The method of claim 1, wherein the radiation comprises an electromagnetic radiation having a wavelength selected from the group of between 200 and 400 nm, between 400 and 760 nm, and between 760 and 3000 nm.

22. The method of claim 1, wherein prior to illumination, the plurality of cells is contacted with a reagent that selectively binds to substantially all of the cells, the reagent capable of generating a signal detectable as a first property of light.

23. A method for purifying one or more selected cells, comprising:

(a) illuminating a population of cells in a frame, wherein the illuminated cells are contained in a plurality of cells immobilized in proximity to a capture matrix, the capture matrix capable of localizing a product secreted by one or more of the cells;

(b) detecting two or more properties of light directed from the frame, wherein a first property of light identifies substantially all cells of the population, and a second property of light identifies product localized to the capture matrix;

(c) locating (i) substantially all cells of the population with reference to the detected first property of light, and (ii) one or more selected cells with reference to the detected second property of light, and

(d) irradiating the non-selected cells, wherein each non-selected cell receives a substantially lethal dose of radiation,

whereby one or more selected cells having a desired product secretion profile are purified.

24. The method of claim 23, wherein the product is selected from a polypeptide, an antibody, an antibody fragment, a cytokine, a growth factor, an enzyme, a hormone, a neurotransmitter, a signaling molecule, and a therapeutic protein.

25. The method of claim 23, wherein the capture matrix comprises a substance selected from Protein G, Protein A, an antibody, an antibody fragment, an aptamer, and a ligand for the product.

26. The method of claim 23, wherein the capture matrix comprises a gel.

27. The method of claim 23, further comprising the step of (e) illuminating a further population of the cells, the further population contained in a further frame, and repeating steps (b) through (d).

28. The method of claim 23 or 27, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the population.

29. The method of claim 23 or 27, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the population.

30. The method of claim 23 or 27, wherein the one or more selected cells are cells that produce an undetectable level of product.

31. The method of claim 27, further comprising repeating step (e) followed by steps (b) through (d) until substantially all of the non-selected cells in the plurality of cells receive a substantially lethal dose of radiation.

32. The method of claim 31, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the plurality.

33. The method of claim 31, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the plurality.

34. The method of claim 31, wherein the one or more selected cells are cells that produce an undetectable level of product.

35. The method of claim 23, further comprising allowing the purified one or more selected cells to proliferate.

36. The method of claim 23, wherein steps (a) through (d) are automated.

37. The method of any of claims 23, 27 or 31, wherein the frame is a portion of a field-of-view, the field-of-view having an area selected from the group of greater than 5 mm<sup>2</sup>, greater than 10 mm<sup>2</sup>, greater than 20 mm<sup>2</sup>, greater than 40 mm<sup>2</sup>, greater than 80 mm<sup>2</sup>, and greater than 160 mm<sup>2</sup>.

38. The method of claim 23, wherein the one or more selected cells are identified with reference to a signal value corresponding to an amount of product localized to the capture matrix in the vicinity of the one or more selected cells.

39. The method of claim 23, wherein the substantially lethal dose of radiation is delivered to each non-selected cell in one or more radiation pulses.

40. The method of claim 23, wherein the substantially lethal dose of radiation has an energy density selected from the group of greater than 0.1 J/cm<sup>2</sup>, greater than 0.3 J/cm<sup>2</sup>, greater than 1 J/cm<sup>2</sup>, greater than 3 J/cm<sup>2</sup>, greater than 10 J/cm<sup>2</sup>, greater than 30 J/cm<sup>2</sup>, and greater than 100 J/cm<sup>2</sup>.

41. The method of claim 23, wherein the substantially lethal dose of radiation has an irradiance selected from the group of greater than 10<sup>7</sup> W/cm<sup>2</sup>, greater than 10<sup>8</sup> W/cm<sup>2</sup>, greater than 10<sup>9</sup> W/cm<sup>2</sup>, greater than 10<sup>10</sup> W/cm<sup>2</sup>, and greater than 10<sup>11</sup> W/cm<sup>2</sup>.

42. The method of claim 23, wherein the radiation comprises an electromagnetic radiation having a wavelength selected from the group of between 200 and 400 nm, between 400 and 760 nm, and between 760 and 3000 nm.

43. The method of claim 23, wherein prior to illuminating, the plurality of cells is contacted with a reagent that binds to substantially all of the cells, the reagent capable of generating a signal detectable as a first property of light.

44. A method for purifying one or more selected cells, comprising:

(a) illuminating a population of cells in a frame, wherein the illuminated cells are contained in a plurality of cells immobilized in proximity to a capture matrix, the capture matrix capable of localizing a product secreted by one or more of the cells;

(b) detecting at least one property of light directed from the frame, wherein a property of light identifies product localized to the capture matrix;

(c) locating (i) one or more selected cells with reference to the detected property of light, and (ii) one or more domains in the frame, each domain corresponding to an area occupied by at least one selected cell, wherein the one or more domains are located with reference to the detected property of light;

(d) irradiating the non-domain area contained in the frame, wherein substantially all cells present within the non-domain area receive a substantially lethal dose of radiation,



whereby one or more selected cells having a desired product secretion profile are purified.

45. The method of claim 44, wherein the product is selected from a polypeptide, an antibody, an antibody fragment, a cytokine, a growth factor, an enzyme, a hormone, a neurotransmitter, a signaling molecule, and a therapeutic protein.

46. The method of claim 44, wherein the capture matrix comprises a substance selected from Protein G, Protein A, an antibody, an antibody fragment, an aptamer, and a ligand for the product.

47. The method of claim 44, wherein the capture matrix comprises a gel.

48. The method of claim 44, further comprising the step of (e) illuminating a further population of the cells, the further population contained in a further frame, and repeating steps (b) through (d).

49. The method of claim 44 or 48, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the population.

50. The method of claim 44 or 48, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the population.

51. The method of claim 44 or 48, wherein the one or more selected cells are cells that produce an undetectable level of product relative to other cells of the population.

52. The method of claim 48, further comprising repeating step (e) followed by steps (b) through (d) until substantially all of the non-selected cells in the plurality of cells receive a substantially lethal dose of radiation.

53. The method of claim 52, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the plurality.

54. The method of claim 52, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the plurality.

55. The method of claim 52, wherein the one or more selected cells are cells that produce an undetectable level of product relative to other cells of the plurality.

56. The method of claim 44, further comprising allowing the purified one or more selected cells to proliferate.

57. The method of claim 44, wherein steps (a) through (d) are automated.

58. The method of any of claims 44, 48 or 52, wherein the frame is a portion of a field-of-view, the field-of-view having an area selected from the group of greater than 5 mm<sup>2</sup>, greater than 10 mm<sup>2</sup>, greater than 20 mm<sup>2</sup>, greater than 40 mm<sup>2</sup>, greater than 80 mm<sup>2</sup>, and greater than 160 mm<sup>2</sup>.

59. The method of claim 44, wherein the one or more selected cells are identified with reference to a signal value corresponding to an amount of product localized to the capture matrix in the vicinity of the one or more selected cells.

60. The method of claim 44, wherein the substantially lethal dose of radiation is delivered in one or more radiation pulses to each cell present within the non-domain area.

61. The method of claim 44, wherein the substantially lethal dose of radiation has an energy density selected from the group of greater than  $0.1 \text{ J/cm}^2$ , greater than  $0.3 \text{ J/cm}^2$ , greater than  $1 \text{ J/cm}^2$ , greater than  $3 \text{ J/cm}^2$ , greater than  $10 \text{ J/cm}^2$ , greater than  $30 \text{ J/cm}^2$ , and greater than  $100 \text{ J/cm}^2$ .

62. The method of claim 44, wherein the substantially lethal dose of radiation has an irradiance selected from the group of greater than  $10^7 \text{ W/cm}^2$ , greater than  $10^8 \text{ W/cm}^2$ , greater than  $10^9 \text{ W/cm}^2$ , greater than  $10^{10} \text{ W/cm}^2$ , and greater than  $10^{11} \text{ W/cm}^2$ .

63. The method of claim 44, wherein the radiation comprises an electromagnetic radiation having a wavelength selected from the group of between 200 and 400 nm, between 400 and 760 nm, and between 760 and 3000 nm.

64. A method for purifying one or more selected cells, comprising:

(a) contacting a plurality of cells immobilized in proximity to a capture matrix, the capture matrix capable of localizing a product secreted by one or more of the cells, with an agent that selectively binds to the product, the agent capable of generating a signal detectable as a property of light;

(b) illuminating a population of the cells, the population contained in a frame;

(c) detecting at least one property of light directed from the frame, wherein a property of light identifies product localized to the capture matrix;

(d) locating (i) one or more selected cells with reference to the detected property of light, and (ii) one or more domains in the frame, each domain corresponding to an area occupied by at least one selected cell, wherein the one or more domains are located with reference to the detected property of light, and

(e) irradiating the non-domain area contained in the frame, wherein each cell present within the non-domain area receives a substantially lethal dose of radiation,

whereby one or more selected cells having a desired product secretion profile are purified.

65. The method of claim 64, wherein the product is selected from a polypeptide, an antibody, an antibody fragment, a cytokine, a growth factor, an enzyme, a hormone, a neurotransmitter, a signaling molecule, and a therapeutic protein.

66. The method of claim 64, wherein the capture matrix comprises a substance selected from Protein G, Protein A, an antibody, an antibody fragment, an aptamer, and a ligand for the product.

67. The method of claim 64, wherein the capture matrix comprises a gel.

68. The method of claim 64, further comprising the step of (f) illuminating a further population of the cells, the further population contained in a further frame, and repeating steps (c) through (e).

69. The method of claim 64 or 68, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the population.

70. The method of claim 64 or 68, wherein the one or more selected cells are cells that produce a low level of product relative to other cells of the population.

71. The method of claim 64 or 68, wherein the one or more selected cells are cells that produce an undetectable level of product relative to other cells of the population.

72. The method of claim 68, further comprising repeating step (f) followed by steps (c) through (e) until substantially all of the non-selected cells in the plurality of cells receive a substantially lethal dose of radiation.

73. The method of claim 72, wherein the one or more selected cells are cells that produce a high level of product relative to other cells of the plurality.

74. The method of claim 72, wherein the one or more selected product-secreting cells are cells that produce a low level of product relative to other cells of the plurality.

75. The method of claim 72, wherein the one or more selected cells are cells that produce an undetectable level of product relative to other cells of the plurality.

76. The method of claim 64, further comprising allowing the purified one or more selected cells to proliferate.

77. The method of claim 64, wherein steps (b) through (e) are automated.

78. The method of any of claims 64, 68 or 72, wherein the frame is a portion of a field-of-view, the field-of-view having an area selected from the group of greater than 5 mm<sup>2</sup>, greater than 10 mm<sup>2</sup>, greater than 20 mm<sup>2</sup>, greater than 40 mm<sup>2</sup>, greater than 80 mm<sup>2</sup>, and greater than 160 mm<sup>2</sup>.

79. The method of claim 64, wherein the agent that selectively binds to the product is selected from an antibody, an antibody fragment, an aptamer, a substrate, and a ligand.

80. The method of claim 64, wherein the one or more selected cells are identified with reference to a signal value corresponding to an amount of product localized to the capture matrix in the vicinity of the one or more selected cells.

81. The method of claim 64, wherein the substantially lethal dose of radiation is delivered in one or more radiation pulses to each cell present within the non-domain area.

82. The method of claim 64, wherein the substantially lethal dose of radiation has an energy density selected from the group of greater than 0.1 J/cm<sup>2</sup>, greater than 0.3 J/cm<sup>2</sup>, greater than 1 J/cm<sup>2</sup>, greater than 3 J/cm<sup>2</sup>, greater than 10 J/cm<sup>2</sup>, greater than 30 J/cm<sup>2</sup>, and greater than 100 J/cm<sup>2</sup>.

83. The method of claim 64, wherein the substantially lethal dose of radiation has an irradiance selected from the group of greater than 10<sup>7</sup> W/cm<sup>2</sup>, greater than 10<sup>8</sup> W/cm<sup>2</sup>, greater than 10<sup>9</sup> W/cm<sup>2</sup>, greater than 10<sup>10</sup> W/cm<sup>2</sup>, and greater than 10<sup>11</sup> W/cm<sup>2</sup>.

84. The method of claim 64, wherein the radiation comprises an electromagnetic radiation having a wavelength selected from the group of between 200 and 400 nm, between 400 and 760 nm, and between 760 and 3000 nm.